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Terms	Documents
L1 and (Phe570 or Phe667)	2

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L2 L1 and (Phe570 or Phe667)
L1 DNA adj polymerase

2 L2
18570 L1

END OF SEARCH HISTORY

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1. Document ID: US 20010012613 A1

L2: Entry 1 of 2

File: PGPB

Aug 9, 2001

PGPUB-DOCUMENT-NUMBER: 20010012613

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20010012613 A1

TITLE: THERMOSTABLE POLYMERASES HAVING ALTERED FIDELITY AND METHOD OF IDENTIFYING AND USING SAME

PUBLICATION-DATE: August 9, 2001

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
LOEB, LAWRENCE A.	BELLEVUE	WA	US	
HOOD, LEROY	SEATTLE	WA	US	
SUZUKI, MOTOSHI	NOGOYA		JP	

US-CL-CURRENT: 435/6
[Full](#) | [Title](#) | [Citation](#) | [Front](#) | [Review](#) | [Classification](#) | [Date](#) | [Reference](#) | [Sequences](#) | [Attachments](#) | [Claims](#) | [KMC](#) | [Draw Desc](#) | [Image](#)

2. Document ID: US 5614365 A

L2: Entry 2 of 2

File: USPT

Mar 25, 1997

US-PAT-NO: 5614365

DOCUMENT-IDENTIFIER: US 5614365 A

TITLE: DNA polymerase having modified nucleotide binding site for DNA sequencing
[Full](#) | [Title](#) | [Citation](#) | [Front](#) | [Review](#) | [Classification](#) | [Date](#) | [Reference](#) | [Sequences](#) | [Attachments](#) | [Claims](#) | [KMC](#) | [Draw Desc](#) | [Image](#)
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Terms	Documents
L1 and (Phe570 or Phe667)	2

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[Previous Page](#) [Next Page](#)

=> d his

(FILE 'HOME' ENTERED AT 09:58:42 ON 21 MAY 2002)

INDEX 'ADISALERTS, ADISINSIGHT, ADISNEWS, AGRICOLA, ANABSTR, AQUASCI,
BIOBUSINESS, BIOCOMMERCE, BIOSIS, BIOTECHABS, BIOTECHDS, BIOTECHNO,
CABA,
CANCERLIT, CAPLUS, CEABA-VTB, CEN, CIN, CONFSCI, CROPB, CROPY, DDFB,
DDFU, DGENE, DRUGB, DRUGLAUNCH, DRUGMONOG2, ...' ENTERED AT 09:58:55 ON
21 MAY 2002

SEA DNA (W) POLYMERASE

246 FILE ADISALERTS
77 FILE ADISINSIGHT
15 FILE ADISNEWS
979 FILE AGRICOLA
49 FILE ANABSTR
209 FILE AQUASCI
243 FILE BIOBUSINESS
265 FILE BIOCOMMERCE
18424 FILE BIOSIS
2695 FILE BIOTECHABS
2695 FILE BIOTECHDS
11411 FILE BIOTECHNO
1310 FILE CABA
8085 FILE CANCERLIT
19438 FILE CAPLUS
291 FILE CEABA-VTB
25 FILE CEN
88 FILE CIN
520 FILE CONFSCI
13 FILE CROPB
12 FILE CROPY
355 FILE DDFB
1086 FILE DDFU
8207 FILE DGENE
355 FILE DRUGB
21 FILE DRUGNL
1628 FILE DRUGU
47 FILE DRUGUPDATES
77 FILE EMBAL
20200 FILE EMBASE
3774 FILE ESBIOWARE
606 FILE FEDRIP
28 FILE FROSTI
89 FILE FSTA
647180 FILE GENBANK
33 FILE HEALSAFE
979 FILE IFIPAT
2630 FILE JICST-EPLUS
4 FILE KOSMET
8942 FILE LIFESCI
2 FILE MEDICONF
21235 FILE MEDLINE
125 FILE NIOSHTIC
173 FILE NTIS
40 FILE OCEAN
7239 FILE PASCAL

365 FILE PHAR
90 FILE PHIN
524 FILE ROMT
13061 FILE SCISEARCH
2 FILE SYNTHLINE
9330 FILE TOXCENTER
16319 FILE USPATFULL
40 FILE USPAT2
1306 FILE WPIDS
1306 FILE WPINDEX
L1 QUE DNA(W) POLYMERASE

FILE 'MEDLINE, EMBASE, BIOSIS, SCISEARCH, BIOTECHNO, CAPLUS' ENTERED AT
10:04:57 ON 21 MAY 2002

L2 13 S L1 AND (PHE570 OR PHE667) OR (F570 OR F667)
L3 6 DUP REM L2 (7 DUPLICATES REMOVED)
L4 13709 S L1 AND (MUTANT OR VARIANT)
L5 0 S L4 AND (PHE570 OR PHE 570)
L6 38154 S L4 AND (PHE667 OR PHE570) OR (570 OR 667)
L7 3 S L4 AND (PHE667 OR PHE570)
L8 1 DUP REM L7 (2 DUPLICATES REMOVED)

=> d 18 ibib ab

L8 ANSWER 1 OF 1 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.DUPLICATE 1
ACCESSION NUMBER: 2000378102 EMBASE
TITLE: *Thermus aquaticus DNA polymerase I*
mutants with altered fidelity. Interacting
mutations in the O-helix.
AUTHOR: Suzuki M.; Yoshida S.; Adman E.T.; Blank A.; Loeb L.A.;
Gottstein J.
CORPORATE SOURCE: L.A. Loeb, J. Gottstein Mem. Cancer Res. Lab., Dept. of
Pathology, University of Washington, Seattle, WA
98195-7705, United States. laloeb@u.washington.edu
SOURCE: Journal of Biological Chemistry, (20 Oct 2000) 275/42
(32728-32735).
Refs: 35
ISSN: 0021-9258 CODEN: JBCHA3
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English
AB **Phe667** in the conserved O-helix of *Thermus aquaticus* (*Taq*)
DNA polymerase I (pol I) is known to be important for
discrimination against dideoxy-NTPs. We show here that **Phe667** is
also important for base selection fidelity. In a forward mutation assay
at high polymerase concentration, wild type pol I catalyzed frequent A
.fwdarw. T and G .fwdarw. T transversions and -1 frameshifts at
nonreiterated sites involving loss of a purine immediately downstream of
a pyrimidine. The **mutants** F667L and A661E,I665T, F667L exhibited
large decreases in A .fwdarw. T and G .fwdarw. T transversions, and the
triple **mutant** displayed reduction in the aforementioned -1
frameshifts as well. Kinetic analysis showed that the F667L and
A661E,I665T, F667L polymerases discriminated against synthesis of A:A
mispairs more effectively and catalyzed less extension of A:A mispairs
than the wild type enzyme. These data indicate that **Phe667**
functions in maintaining the error frequency and spectrum, and the
catalytic efficiency, of wild type pol I. We also found that the strong
general mutator activity conferred by the single A661E substitution was
entirely suppressed in the A661E, I665T,F667L polymerase, exemplifying
how interactions among O-helix residues can contribute to fidelity. We
discuss the mutator and anti-mutator mutations in light of recently obtained
three-dimensional structures of *T. aquaticus* pol I.

=> d 13 ibib ab 1-6

L3 ANSWER 1 OF 6 CAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 2000:260305 CAPLUS
DOCUMENT NUMBER: 132:265445
TITLE: Preparation of nucleotide compounds including a rigid linker used in DNA sequencing
INVENTOR(S): Kahn, Shaheer H.; Rosenblum, Barnett B.; Zhen, Weiguo; Menchen, Steven M.
PATENT ASSIGNEE(S): The Perkin-Elmer Corporation, USA
SOURCE: PCT Int. Appl., 63 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2000021974	A1	20000420	WO 1999-US12323	19990602
W: AU, CA, JP				
RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
US 6096875	A	20000801	US 1998-172789	19981014
AU 9946740	A1	20000501	AU 1999-46740	19990602
EP 1121371	A1	20010808	EP 1999-930140	19990602
R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
PRIORITY APPLN. INFO.:			US 1998-172789	A 19981014
			WO 1999-US12323	W 19990602

OTHER SOURCE(S): MARPAT 132:265445
AB A nucleoside/tide compd. having a rigid linker attached to the 8-position of a purine, the 7-position of a 7-deazapurine and the 5-position of a pyrimidine is disclosed. Fluorescent dyes may be attached to this linker and the fluorescent nucleotide used in primer extension reactions. Thus, the fluorescein dye HEX-1 was attached to the 5-position of ddCTP via an acetylene-phenyl-oxyethyleneamino linkage. This nucleotide deriv. was used in DNA sequencing with Taq polymerase contg. an R660S mutation. The synthesis of a no. of nucleoside/nucleotide derivs. contg. various rigid linkers is described.

REFERENCE COUNT: 5 THERE ARE 5 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE REFORMAT

L3 ANSWER 2 OF 6 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.DUPLICATE 1
ACCESSION NUMBER: 2000378102 EMBASE
TITLE: *Thermus aquaticus DNA polymerase I mutants with altered fidelity. Interacting mutations in the O-helix.*
AUTHOR: Suzuki M.; Yoshida S.; Adman E.T.; Blank A.; Loeb L.A.; Gottstein J.
CORPORATE SOURCE: L.A. Loeb, J. Gottstein Mem. Cancer Res. Lab., Dept. of Pathology, University of Washington, Seattle, WA 98195-7705, United States. laloeb@u.washington.edu
SOURCE: Journal of Biological Chemistry, (20 Oct 2000) 275/42 (32728-32735).

Refs: 35
ISSN 0021-9258 CODEN: JBCHA3
COUNTRY: United States
DOCUMENT TYPE: Journal; Article
FILE SEGMENT: 029 Clinical Biochemistry
LANGUAGE: English
SUMMARY LANGUAGE: English
AB Phe667 in the conserved O-helix of Thermus aquaticus (Taq) DNA polymerase I (pol I) is known to be important for discrimination against dideoxy-NTPs. We show here that Phe667 is also important for base selection fidelity. In a forward mutation assay at high polymerase concentration, wild type pol I catalyzed frequent A .fwdarw. T and G .fwdarw. T transversions and -1 frameshifts at nonreiterated sites involving loss of a purine immediately downstream of a pyrimidine. The mutants F667L and A661E, I665T, F667L exhibited large decreases in A .fwdarw. T and G .fwdarw. T transversions, and the triple mutant displayed reduction in the aforementioned -1 frameshifts as well. Kinetic analysis showed that the F667L and A661E, I665T, F667L polymerases discriminated against synthesis of A:A mispairs more effectively and catalyzed less extension of A:A mispairs than the wild type enzyme. These data indicate that Phe667 functions in maintaining the error frequency and spectrum, and the catalytic efficiency, of wild type pol I. We also found that the strong general mutator activity conferred by the single A661E substitution was entirely suppressed in the A661E, I665T, F667L polymerase, exemplifying how interactions among O-helix residues can contribute to fidelity. We discuss the mutator and anti-mutator mutations in light of recently obtained three-dimensional structures of T. aquaticus pol I.

L3 ANSWER 3 OF 6 CAPLUS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1999:571772 CAPLUS
DOCUMENT NUMBER: 131:196102
TITLE: Nucleotide compounds including a rigid linker
INVENTOR(S): Khan, Shaheer H.; Rosenblum, Barnett B.; Zhen, Weiguo;
Menchen, Steven M.
PATENT ASSIGNEE(S): USA
SOURCE: U.S., 32 pp.
CODEN: USXXAM
DOCUMENT TYPE: Patent
LANGUAGE: English
FAMILY ACC. NUM. COUNT: 2
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
US 5948648	A	19990907	US 1998-87250	19980529
US 6197555	B1	20010306	US 1999-461510	19991214
PRIORITY APPLN. INFO.:			US 1998-87250	A2 19980529
			US 1998-172789	A3 19981014

OTHER SOURCE(S): MARPAT 131:196102
AB A nucleoside/tide compd. having a rigid linker attached to the 8-position of a purine, the 7-position of a 7-deazapurine and the 5-position of a pyrimidine is disclosed. Fluorescent dyes may be attached to this linker and the fluorescent nucleotide used in primer extension reactions. Thus, the fluorescein dye HEX-1 was attached to the 5-position of ddCTP via an acetylene-phenyl-oxyethyleneamino linkage. This nucleotide deriv. was used in DNA sequencing with Taq polymerase contg. an R660S mutation. The synthesis of a no. of nucleoside/nucleotide derivs. contg. various rigid linkers is described.

REFERENCE COUNT: 7 THERE ARE 7 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE REFORMAT

L3 ANSWER 4 OF 6

MEDLINE

DUPLICATE 2

ACCESSION NUMBER: 19907133 MEDLINE
DOCUMENT NUMBER: 9907133 PubMed ID: 10101201
TITLE: Dye structure affects Taq DNA polymerase terminator selectivity.
AUTHOR: Brandis J W
CORPORATE SOURCE: DNA Chemistry Group, Genetic Analysis Business Unit, PE Biosystems, 850 Lincoln Center Drive, Foster City, CA 94404, USA.. brandjw@perkin-elmer.com
SOURCE: NUCLEIC ACIDS RESEARCH, (1999 Apr 15) 27 (8) 1912-8.
PUB. COUNTRY: ENGLAND: United Kingdom
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 199906
ENTRY DATE: Entered STN: 19990628
Last Updated on STN: 19990628
Entered Medline: 19990614

AB All DNA sequencing methods have benefited from the development of new F667Y versions of Taq DNA polymerase. However, terminator chemistry methods show less uniform peak height patterns when compared to primer chemistry profiles suggesting that the dyes and/or their linker arms affect enzyme selectivity. We have measured elementary nucleotide rate

and binding constants for representative rhodamine- and fluorescein-labeled terminators to determine how they interact with F667 versions of Taq Pol I. We have also developed a rapid gel-based selectivity assay

that can be used to screen and to quantify dye-enzyme interactions with F667Y versions of the enzyme. Our results show that 6-TAMRA-ddTTP behaves like unlabeled ddTTP, while 6-FAM-ddTTP shows a 40-fold reduction in the rate constant for polymerization without affecting ground-state nucleotide binding. Detailed mechanism studies indicate that both isomers of different fluorescein dyes interfere with a conformational change step which the polymerase undergoes following nucleotide binding but only when these dyes are attached to pyrimidines. When these same dyes are attached to purines by the same propargylamino linker arm, they show no effect on enzyme selectivity. These studies suggest that it may be possible to develop fluorescein terminators for thermocycle DNA sequencing methods

for polymerases that do not discriminate between deoxy- and dideoxynucleotides.

L3 ANSWER 5 OF 6 CAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1992:423036 CAPLUS
DOCUMENT NUMBER: 117:23036
TITLE: Regulation of tubercidin biosynthesis in Streptomyces tubercidicus by adenine and histidine
AUTHOR(S): Yoo, Jin Cheol; Hah, Yung Chil
CORPORATE SOURCE: Dep. Pharm., Chosun Univ., Kwangju, 501-759, S. Korea
SOURCE: Misaengmul Hakhoechi (1991), 29(3), 160-6
DOCUMENT TYPE: CODEN: MIHCAR; ISSN: 0440-2413
LANGUAGE: Journal
Korean

AB The regulatory mechanism of tubercidin biosynthesis in S. tubercidicus was

studied. In a wide-type strain, addn. of adenine and histidine into the medium decreased the tubercidin prodn. by 60-65% and 40%, resp. The effects of adenine and histidine were alleviated by the addn. of inosine monophosphate and 5-aminoimidazole-4-carboxamide ribotide. The prodn. of tubercidin in S. tubercidicus strain K115 (ade-) was nearly shut off by histidine. In contrast to strain K115, adenine inhibited tubercidin biosynthesis in S. tubercidicus strain K412 (his-). In S. tubercidicus F667 strain (ade-, his-), tubercidin prodn. was increased by adenine and histidine.

L3 ANSWER 6 OF 6 CACIS COPYRIGHT 2002 ACS
ACCESSION NUMBER: 1967:26509 CAPLUS
DOCUMENT NUMBER: 66:26509
TITLE: A mathematical method for the quantitative determination of proline and citrulline by automatic amino acid analysis
AUTHOR(S): Breuer, Josef; Ise, H.; Doellefeld, Erich; Breuer, Heinz
CORPORATE SOURCE: Abt. Klin. Chem. Biochem. Chirurgischen Univ. Poliklin., Bonn, Ger.
SOURCE: Z. Klin. Chem. (1966), 4(5), 267-8
CODEN: ZKLCAY
DOCUMENT TYPE: Journal
LANGUAGE: German
AB By using an automatic amino acid analyzer, it is possible to resolve mixts. of proline (I) and citrulline (II) by application of a math. method. The method is based on the differences in the absorption curves of the reaction products of ninhydrin and I and II, i.e., the I product has a broader absorption peak at 440 m.mu. than at 570 m.mu., while the reverse is true for II product. With norleucine as an internal standard, the following formulas were derived for the quant. detns.: I (mg.) = $(F440(KC570) - F570(KC440))/(KP440(KC570) - KP570(KC440))$ and II (mg.) = $(F440(KP570) - F570(KP440))/(KC440 - (KP570 - KC570(KP440)))$, where F is the ratio of the area under the denoted wavelength absorption curve for test mixt. to the curve area for standard norleucine at 570 m.mu., and KC and KP are the curve areas for pure II and I, resp., at the denoted wavelengths.